

Variation in Elemental Content of Lettuce
Grown Under Base-Line Conditions in Five
Controlled-Environment Facilities

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Variation in Elemental Content of Lettuce Grown under Base-line Conditions in Five Controlled-environment Facilities¹

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Abstract. An interlaboratory comparison was made of the variation in elemental concentration of leaf lettuce (*Lactuca sativa* L. cv. Grand Rapids) plants grown under base-line conditions for 28 days in 5 controlled-environment facilities. Two studies were conducted by each of 5 investigators using a sphagnum peat-vermiculite mix obtained from a common source. Plant tissue from all studies was collected and analyzed at 1 laboratory for 10 essential elements (P, K, Ca, Mg, Fe, Mn, B, Zn, Cu, Mo) and 10 non-essential elements (Al, Si, Ti, Sr, Ba, Na, Pb, V, Li, and Sn) by arc emission spectroscopy. The 10 essential elements occurred at concentrations adequate for normal lettuce growth. Tissue analysis data showed large differences in elemental concentration among experiments conducted by the same investigator as well as by different investigators suggesting inadvertent contamination of the leaf samples by the rooting media or contamination of the watering system in each controlled-environment facility. These differences, however, had no observable effect on vegetative growth. Based on a nested analysis on variance, 95% of the elements showed significant plant to plant differences in concentration. However, based on an analysis of variance components, the greatest source of variation was found among investigators.

Since 1969 the American Society of Horticultural Science Working Group on Growth Chambers and Controlled Environments has attempted to standardize growing and reporting procedures for plant growth chamber experiments (2, 3, 13, 21, 22, and 23). Many investigators (15, 16) have analyzed vegetative growth in controlled environments, but few have quantified variability within or among growth chambers (11, 24, 35) and tissue analysis in such studies is uncommon (17, 18, 19, 22, 26).

We report herein the comparative concentration of 10 essential and 10 non-essential elements found in leaf tissue of 28-year-old lettuce grown under standard base-line conditions. We further evaluate the variation in concentration of these elements as-

sociated with analytical procedures, plant-to-plant differences within a single experiment, differences between experiments for a given investigator, and differences among investigators.

Materials and Methods

Plant materials. 'Grand Rapids' lettuce seed from a single seed lot⁸ was stored at the Plant Stress Laboratory in Beltsville, Md., and was distributed to each experimenter as needed.

Experimental conditions. We grew lettuce plants using standard environmental conditions (13) with growing and environmental measurement procedures standardized to minimize variability and inadvertent stress (21, 32, 33). To assure uniformity of experimental conditions, a set of calibrated environmental measuring instruments was circulated among investigators for each study. Plants were grown under environmental conditions, specified by the working group, and found previously (13) to produce uniform, normal-appearing lettuce plants of good yield; no attempt was made to maximize yield. Peat-vermiculite mix was used as the standard growth medium.

The studies were conducted during 1973-1975 in the 5 laboratories indicated in Table 1. Each investigator conducted 2 studies (Table 1). The studies were conducted in the same facility within each laboratory to avoid growth chamber differences as a source of variation. A detailed description of environmental facilities in each laboratory was provided by Hammer et al. (13), Tibbitts et al. (34), Krizek and Berry (19), and Krizek and Ormrod (20).

Environmental conditions established in each facility are summarized in Table 2. The plants were grown from seed on a 16hr photoperiod with 1500 ma Cool White fluorescent and incandescent lamps. These lamps provided approximately 325 $\mu\text{E m}^{-2}\text{s}^{-1}$ of photosynthetically active radiation (PAR) as measured with a LI-COR¹ quantum flux sensor (sensitivity between 400-700 nm). An air temperature of 25°C day and 20° night was provided. Relative humidity was controlled where possible at 70% both day and

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Table 1. Investigators, laboratories, and chamber specifications in the different laboratories.

Code	Investigator	Location	Chamber manufacturer and type	Barrier material	Reflective specifications of surfaces
A	J. C. McFarlane	Env. Monitoring Sys. Lab., EPA Las Vegas, Nevada	Environmental Growth Chamber M-3, Reach-in	Glass	Polished stainless steel
B	D. T. Krizek	Plant Stress Lab., USDA Beltsville, Maryland	Environmental Growth Chamber M-3, Reach-in	Plexiglass	Polished stainless steel
C	T. W. Tibbitts	Biotron, Univ. of Wisconsin Madison, Wisconsin	Built-in growth rooms	Plexiglass	White paint
D	R. W. Langhans	Post Lab., Cornell Univ., Ithaca, N.Y.	Environmental Growth Chamber, Walk-in	Plexiglass	Aluminized mylar
E	D. P. Ormrod	Hort. Science, Univ. of Guelph, Guelph Ontario, Canada	Controlled Environment Ltd., Reach-in	None	White enamel

^aTransparent barrier between lamps and plant chamber, if any.

Table 2. Environmental conditions measured in the different studies. Laboratories are identified in Table 1.

Lab.	Expt. No.	Radiation at start of study			Air		Temp (°C)		Dew point	
		PAR ^a (μEm ⁻² s ⁻¹)	FR ^b (Wm ⁻² nm ⁻¹)	Total ^b (Wm ⁻²)	Day ^a	Night ^a	Media (day)		Day ^a	Night ^a
		400-700 nm	720-820 nm	280-2800 nm			1 cm depth ^c	6 cm depth ^c		
A	1	315-330	.085	177	24.2-26.2	19.0-21.0	22.0-26.4	23.8-27.8	--	--
	2	312-325	.081	134	24.0-26.0	19.0-21.0	--	--	(19.0) ^d	(14.0) ^d
B	1	281-316	.060	135	24.1-25.3	18.9-20.8	--	--	19.4-22.2	--
	2	300-320	.076	159	25.2-26.3	19.7-21.0	24.8-25.3	--	--	15.5-16.8
C	1	347-355	.088	140	25.0-26.0	19.0-22.0	24.3-25.5	23.9-24.6	16.9-19.7	14.2-17.2
	2	345-360	.083	135	24.5-25.8	18.5-21.3	24.4-25.5	23.3-24.6	15.8-19.0	13.4-17.3
D	1	320-335	.082	133	24.0-26.0	19.0-21.0	25.7-26.1	25.7-25.9	18.0-20.0	15.0-17.0
	2	325-330	.082	138	24.0-26.0	19.0-21.0	25.7-26.1	25.7-25.9	18.0-20.0	15.0-17.0
E	1	332-346	.067	161	24.0-27.0	19.0-21.0	25.2-25.8	26.0-26.8	18.0-22.0	13.0-17.5
	2	328-346	.076	164	25.0-28.0	20.0-21.0	26.1-26.6	27.0-27.5	19.0-22.0	15.0-17.4

^aRange over the growing area at the start of the study.

^bMeasured over the center container at the start of the study.

^cRange of avg daily temp. during the study.

^dRange between the containers at the start of the study.

night. Ambient CO₂ level was used with make-up air provided in all chambers.

Cultural procedures. The plants were seeded, selected for uniformity, and grown in 1-liter white plastic pots containing a peat-vermiculite mix (1 sphagnum peat: 1 vermiculite v/v with added nutrients) obtained from a common source and distributed to each cooperator. The plants were grown for 28 days in accordance with previously described base-line cultural procedures. Plants were irrigated by an automatic watering system every 4 hr from day 0 to 21 with 50 ml of nutrient solution and from day 22 to 28 with 100 ml per pot at the rate of 20 to 50 ml per min. This volume provided at least 25 ml of drainage at each watering, thus assuring a relatively constant water content close to field capacity and uniform nutrient availability without salt build-up.

Nutrient solution. A modified Hoagland solution was prepared using reagent-grade chemicals and distilled or demineralized water (13). The macronutrients in the nutrient solution in mmol liter⁻¹ were 7.5 NO₃⁻, 0.5 H₂PO₄⁻, 3K⁺, 2.5 Ca⁺⁺, 1 Mg⁺⁺, 1 SO₄⁻, 0.5 Na⁺ and 0.5 Cl.

Micronutrients in μmol liter⁻¹ were 23 B, 4.6 Mn, 0.38 Zn, 0.15 Cu, and 0.05 Mo. Iron at 44 μmol liter⁻¹ was added as the ethy-

lenediaminetetraacetate (EDTA) complex (13).

Experimental design and tissue analysis. In each experiment, 20 plants were grown; 5 were harvested for growth measurements at weekly intervals, beginning on day 7. After each harvest, the remaining plants were rerandomized as to location in the growth chamber. The leaf tissue of the 5 plants from the last harvest (28 days) was used for tissue analysis. Leaves were dried in a forced draft oven at 80°C for 2 days. After determination of dry weight, each plant was ground with a mortar and pestle and placed in a sample vial. Samples were forwarded to a central laboratory (W. L. Berry) for analysis by arc emission spectrography (36). Comparative analyses were run on orchard leaf tissue obtained from the National Bureau of Standards (NBS) to check for accuracy of the analytical methods. Tissue was analyzed and reported for P, K, Ca, Mg, Fe, Mn, B, Zn, Cu, Mo, Al, Si, Ti, Sr, Ba, Na, Pb, V, Sn, and Li. Analyses were also performed for Co, Ni, Cr, Ag, and Cd, but concentrations were less than 2 μgg⁻¹ (2 ppm). Five elemental determinations were made on 5 separate sub-samples of the tissue from each plant (N×5).

Statistical analysis. The data were analyzed by nested analysis of variance (Table 4). The components of variance contributed by

Table 3. Comparative growth of 'Grand Rapids' lettuce treated for 28 days from seeding under base-line conditions. Data for Investigators A, C, and D, were taken from Hammer *et al.*, (13) and for B from Krizek and Ormrod (19).

Growth parameter	Investigator					
	B		A, C, D		E	
	Mean ^a	Range	Mean ^a	Range	Mean ^a	Range
Cotyledon length ^b (mm)	22.0	20.4–23.4	22.6	(21–26)	23.6	22.6–25.4
Cotyledon width ^b (mm)	11.0	10.6–11.4	11.7	(9–14)	12.2	12.0–12.6
No. of nodes with leaf 1 cm long	18.0	16.6–19.6	18.8	16–23	21.5	19–23
Length of the 5th leaf (mm)	157.3	147.8–163.0	153.8	139–180	157.0	155–159
Width of the 5th leaf	138.4	125.8–151.8	153.8	125–183	171.2	165–181
Stem length ^a (mm)	28.7	23.8–35.0	24.9	16–29	37.3	26–43
Fresh weight of tops (g)	78.5	67.2–92.4	90.4	48.4–119.8	126.5	103.6–138.2
Dry wt of tops (g)	4.0	3.4–4.6	4.7	3.0–6.0	6.8	5.2–7.9

^aMean of the 5 plants in each study.

^bAt 14 days from seeding.

^cFrom cotyledonary node to top node with leaf over 1 cm long.

investigator, experiments within an investigator (e), plant to plant variation within an experiment (p), and variation within subsamples (a) were computed for each element. The ratio of each component to the sum of the variances was calculated and presented as percentage of the total variance for each element.

Results and Discussion

Overall results. The mean concentration of the 10 essential and 10 non-essential elements for the 2 studies conducted by each of the 5 investigators is shown in Tables 5 and 6. The standard error of the means shown are based on the overall variability and include variation contributed by each of the components indicated in Table 4.

Variance components. Investigator variance in Mn, Mo, and Li concentrations accounted for more than half of the total variance for those elements. Only for Ca, Pb, and V did the investigator variance account for less than 25% of the total variance (Table 7 and 8). The variance contributed by differences between experiments for any given investigator accounted for more than 25% of the variance in the case of Mg, Sr, Ba, Pb and Sn and less for all other elements.

Plant to plant variance contributed more than 25% of the overall variance for P, Fe, Cu, Al, Si, Ti, and V. The variance contributed by plants within an experiment was less than 25% for the rest of the listed elements. The variance contributed by analytical procedures accounted for more than 50% of the total variance in the case of K and Na and less than 25% for P, Mg, Mn, B, Cu, Al, Si, Sr, Ba, V, Li, and Sn.

Variability among investigators. The variability in elemental content among investigators was generally ± 5 to 10%. The concentration of certain elements e.g., Fe, Zn, and Mn deviated considerably more than this in studies of some investigators (Table 5, 6). Except for Ca, Mg, B and several non-essential elements, Sr, Ba, Pb, V, and Li, these differences were highly significant.

Table 4. Format for nested analysis of variance of elemental concentration of 'Grand Rapids' lettuce leaves. Study involved 5 investigators, 2 experiments by each investigator, 5 plants per experiment and 5 analytical subsamples per plant.

Source of variance	Degrees of freedom	Expected mean square
Total	249	
Investigator	4	$\sigma_a^2 + 5\sigma_p^2 + 25\sigma_e^2 + 50\sigma^2$
Expts. (e) by investigator	5	$\sigma_a^2 + 5\sigma_p^2 + 25\sigma_e^2$
Plants (p) within expt.	40	$\sigma_a^2 + 5\sigma_p^2$
Subsample within plants (a)	200	σ_a^2

In general, there was a greater amount of Fe, Mn, Zn, Cu, Mo, Al, Si, Ti, Pb, and Li, and lower Na in plants from experiments of Investigator E than in those of the other Investigators (Table 5, 6). This increase may have resulted from contamination of the leaf samples by the peat-vermiculite mix since most of these elements are high in this media. Also, the high Cu in lettuce from Investigator E could be attributed to the use of a Cu pipe in the watering system. High concentration of Mn and low concentration of Fe, Ti, and Li were found in lettuce tissue grown in studies of Investigator D for unknown reasons.

There was a significantly lower amount of K, Mn, Sn and Pb, and a higher amount of Ca, B, Mo, and Na in plants for Investigator A than in plants for some of the other investigators. The relatively low amount of Pb and Sn in lettuce grown by Investigator A may have resulted from the extensive use of plastic piping in that facility. Sodium content was significantly higher in lettuce grown by Investigators A and B than in those of the other investigators, for no apparent reason. The essential elements, Fe and B, and the nonessential elements Al, Si, Ti, and Li tended to be lower for lettuce grown by Investigator D, and generally higher for that grown by Investigator E than for the other investigators. This indicates that a source of contamination, possibly the media, was effectively minimized by Investigator D but not controlled by Investigator E. The Mg concentration of plant tissue from the different laboratories closely paralleled the Ca content of the tissue.

The differences in elemental concentration of plant tissue among individual investigators at different locations were influenced apparently by at least 2 factors. One was the inherent background concentration of nutrients in the water, atmosphere, and general environment which could be especially significant for micronutrient determinations. Differences were especially large for the nonessential elements. The other factor was the degree of quality control that the investigator was able to maintain in following prescribed procedures.

Variability between experiments. The large variability between experiments of any single investigator was demonstrated by the magnitude of the standard error of the mean in Tables 5 and 6, and by the components of variance in Tables 7 and 8. Except for P, Fe, Cu, Mo, Al, Si, Ti, Na, and V there were significant differences in variation for all elements (Tables 5 and 6) between experiments within a single laboratory.

The difference in tissue concentration between experiments for most elements at any 1 location was often so large that it tended to obscure differences that may have been present among locations.

Table 5. Tissue concentration of 10 essential elements in leaves of 'Grand Rapids' lettuce plants grown under base-line conditions in controlled environments for 28 days from seeding. Results of nested analysis of variance for each element are shown.

Investigator	Elemental concn based on dry wt				
	P (%)	K (%)	Ca (%)	Mg (%)	Fe ($\mu\text{g g}^{-1}$)
A	0.76 $\pm$ 0.11	8.23 $\pm$ 2.8	1.70 $\pm$ 0.21	0.33 $\pm$ 0.04	209 $\pm$ 86
B	0.69 $\pm$ 0.09	9.74 $\pm$ 2.0	1.51 $\pm$ 0.14	0.31 $\pm$ 0.03	192 $\pm$ 84
C	0.55 $\pm$ 0.08	10.00 $\pm$ 2.1	1.23 $\pm$ 0.16	0.27 $\pm$ 0.03	204 $\pm$ 64
D	0.69 $\pm$ 0.12	12.71 $\pm$ 1.5	1.53 $\pm$ 0.39	0.32 $\pm$ 0.06	126 $\pm$ 22
E	0.55 $\pm$ 0.09	10.51 $\pm$ 2.7	1.41 $\pm$ 0.15	0.39 $\pm$ 0.04	276 $\pm$ 86
Mean	0.65 $\pm$ 0.13	10.40 $\pm$ 2.7	1.47 $\pm$ 0.28	0.32 $\pm$ 0.05	202 $\pm$ 87
F test					
Investigator	*	*	NS	NS	*
Expt. by investigator	NS	**	**	**	NS
Plants within expt.	**	NS	**	**	**
Investigator	Mn ($\mu\text{g g}^{-1}$)	B ($\mu\text{g g}^{-1}$)	Zn ($\mu\text{g g}^{-1}$)	Cu ($\mu\text{g g}^{-1}$)	Mo ($\mu\text{g g}^{-1}$)
A	14.4 $\pm$ 2.4	28.8 $\pm$ 7.2	38.6 $\pm$ 9.7	7.4 $\pm$ 1.5	2.7 $\pm$ 0.8
B	45.3 $\pm$ 10.8	21.1 $\pm$ 4.0	63.7 $\pm$ 10.8	12.6 $\pm$ 3.7	1.8 $\pm$ 0.4
C	102.0 $\pm$ 15.9	19.3 $\pm$ 2.0	49.3 $\pm$ 10.1	6.0 $\pm$ 1.3	1.3 $\pm$ 0.3
D	106.5 $\pm$ 19.7	16.2 $\pm$ 2.3	47.1 $\pm$ 14.6	7.0 $\pm$ 2.1	1.2 $\pm$ 0.4
E	135.9 $\pm$ 23.3	21.8 $\pm$ 2.3	71.0 $\pm$ 17.6	13.8 $\pm$ 6.2	1.2 $\pm$ 0.2
Mean	80.8 $\pm$ 47.2	21.4 $\pm$ 5.8	53.9 $\pm$ 17.4	9.4 $\pm$ 4.7	1.6 $\pm$ 0.7
F test					
Investigator	**	NS	*	*	**
Expt. by investigator	**	**	**	NS	NS
Plants within expt.	**	**	**	**	**

*,**,NS Significant at 5% (*), or 1% (**) level or nonsignificant (NS) at 5% level, respectively.

^aMeans and SE of the mean based on 2 studies per investigator, 5 plants per study, and 5 subsamples per plant using N = 50.

^bSE based on variability among investigators using N = 250.

Table 6. Tissue concentration of 10 non-essential elements in leaves of 'Grand Rapids' lettuce plants grown under base-line conditions in controlled environments for 28 days from seeding. Results of nested analysis of variance for each element are shown.

Investigator	Elemental concn based on dry wt				
	Al ($\mu\text{g g}^{-1}$)	Si ($\mu\text{g g}^{-1}$)	Ti ($\mu\text{g g}^{-1}$)	Sr ($\mu\text{g g}^{-1}$)	Ba ($\mu\text{g g}^{-1}$)
A	107 $\pm$ 61	311 $\pm$ 167	19.4 $\pm$ 12.1	15.2 $\pm$ 2.1	21.8 $\pm$ 7.1
B	79 $\pm$ 42	220 $\pm$ 107	21.1 $\pm$ 13.1	13.1 $\pm$ 2.0	20.4 $\pm$ 9.1
C	73 $\pm$ 32	226 $\pm$ 113	18.9 $\pm$ 11.0	10.5 $\pm$ 4.4	11.8 $\pm$ 3.3
D	28 $\pm$ 8	92 $\pm$ 33	3.8 $\pm$ 2.9	16.4 $\pm$ 5.9	8.1 $\pm$ 2.3
E	221 $\pm$ 168	449 $\pm$ 194	28.9 $\pm$ 11.9	20.7 $\pm$ 2.4	17.8 $\pm$ 2.9
Mean	102 $\pm$ 105	259 $\pm$ 178	14.4 $\pm$ 13.5	15.2 $\pm$ 5.0	16.0 $\pm$ 7.6
F test					
Investigator	*	*	**	NS	NS
Expt. by investigator	NS	NS	NS	**	**
Plants within expt.	**	**	**	**	**
Investigator	Na (%)	Pb ($\mu\text{g g}^{-1}$)	V ($\mu\text{g g}^{-1}$)	Sn ($\mu\text{g g}^{-1}$)	Li ($\mu\text{g g}^{-1}$)
A	0.40 $\pm$ 0.12	0.6 $\pm$ 2.1	0.1 $\pm$ 0.4	0.2 $\pm$ 0.7	2.0 $\pm$ 0.7
B	0.33 $\pm$ 0.11	9.9 $\pm$ 8.9	4.3 $\pm$ 9.5	3.1 $\pm$ 2.5	2.1 $\pm$ 1.6
C	0.21 $\pm$ 0.13	9.7 $\pm$ 13.3	1.3 $\pm$ 1.4	5.4 $\pm$ 2.0	1.6 $\pm$ 1.3
D	0.23 $\pm$ 0.10	11.7 $\pm$ 7.6	2.6 $\pm$ 3.1	4.5 $\pm$ 1.4	0.5 $\pm$ 0.7
E	0.16 $\pm$ 0.19	16.2 $\pm$ 5.5	3.1 $\pm$ 1.7	4.7 $\pm$ 1.7	6.4 $\pm$ 1.8
Mean	0.27 $\pm$ 0.16	9.6 $\pm$ 9.7	2.3 $\pm$ 4.8	3.6 $\pm$ 2.5	2.5 $\pm$ 2.4
F test					
Investigator	**	NS	NS	*	NS
Expt. by investigator	NS	**	NS	**	**
Plants within expt.	*	*	**	**	**

*,**,NS Significant at 5% (*) or 1% (**) or nonsignificant (NS) at 5% level, respectively.

^aMeans and SE of the mean based on 2 studies per investigator, 5 plants per study, and 5 subsamples per plant using N = 50.

^bSE based on variability among investigators using N = 250.

Variability among plants. Except for K there were significant differences in plant to plant variation for every element (Table 5 and 6). These differences may have been related to: 1) contamination

associated with experimental procedures, and 2) genetic differences between plants influencing nutrient concentration (1, 10, 27, 29, 31). The latter explanation does not seem likely for lettuce

Table 7. Variance components as percent of the sum of components for 10 elements in lettuce leaves using nested analysis of variance described in Table 4.

Element	Source of total variance			
	Investigator (%)	Expts. by investigator (%)	Plants within expt. (%)	Subsample within plants (%)
P	38	9	30	23
K	28	8	2	66
Ca	22	23	16	39
Mg	36	29	14	21
Fe	29	3	36	32
Mn	88	5	3	4
B	44	23	11	22
Zn	44	10	18	28
Cu	47	2	41	10
Mo	63	0	7	31

Table 8. Variance components as percent of the sum of the components for 10 non-essential elements in lettuce leaves using nested analysis of variance described in Table 4.

Element	Source of total variance			
	Investigator (%)	Expts. by investigator (%)	Plants within expt. (%)	Subsample within plants (%)
Al	37	2	55	6
Si	45	3	29	24
Ti	39	2	33	30
Sr	26	46	21	7
Ba	27	47	15	11
Na	33	1	8	58
Pb	12	38	4	46
V	5	17	66	12
Li	66	17	5	12
Sn	43	29	5	24

since this species is primarily self-pollinated. If there were genetic differences present, only a few individuals with genetic differences in elemental accumulation would be expected to be present in a sample of this size.

Variability among subsamples. A measure of precision of the analytical method for each of the essential elements was obtained through comparative analysis of a sample of standard orchard leaf tissue obtained from the National Bureau of Standards (NBS) and

representative lettuce leaves from this study (the first plant of the first experiment of Investigator C) (Table 9). The agreement with the NBS values for individual elements in orchard leaves was good except for P, which analyzed about 14% low. These values demonstrate the magnitude of variation for individual analyses and illustrate that the variation for certain elements is greater than for others.

The major components of analytical error in the arc emission spectrographic procedure are: 1) the inherent variability of spectrographic analysis; 2) a tissue concentration less than the optimal analytical range; 3) small sample size (37), and 4) heterogeneity of samples.

The analytical variation in the K, P, and Zn analyses could be a contributing factor for observed variation of these elements in the different studies. The concentration of Zn and Mo present in the NBS orchard leaves was on the low end of the analytical curve resulting in a large coefficient of variation (Table 9). The concentration of the nonessential elements, Co, Ni, Cr, Ag, and Cd was below the minimum detectable range ($2\mu\text{gg}^{-1}$) of the arc spectrograph (36).

Some of the variability encountered in the analysis of Ca, Fe, Zn and Cu could have been due to the heterogeneity of the small multi-tissue plant samples, even though they were well mixed before subsampling. Lamb (23), Berry (6) and others have shown that different tissues within the plant may have different concentrations of the same element. For example, in lettuce (6) the petioles and blades differ two-fold in Zn concentration. Since only 6 to 10 mg of dry plant material was used for each individual analysis, some variability can also be expected due to varying percentages of tissues from different plant parts. Increased sample variation would be observed as the result of contamination from small particulate contaminants not uniformly dispersed in the sample.

LaBastide and Van Goor (22) reported significant differences in chemical analyses of leaf tissue among laboratories and within a laboratory even when identical pairs of samples were analyzed. Considering the large degree of variability in tissue analysis reported for identical samples (22), we have clearly demonstrated the value of running chemical analyses in a common laboratory. Had we not done so, it is likely that the differences obtained would have been even greater in magnitude. The high concentrations of Fe, Mn, Al, Si, and Ti found in plants grown in the peat-lite mix probably originated from contamination of the leaves by the mix. We feel that the increased concentration of Ba and Sr in

Table 9. Comparative coefficient of variation (CV) obtained in determining elemental content of standard leaf tissue and a representative lettuce plant from the study.

Element	Orchard leaves ²			Lettuce plant ³	
	NBS value	Determined	CV (%)	Concn	CV (%)
P	(%)	(%)		(%)	
	0.21	0.18	12.5	0.57	10.6
K	1.47	1.48	12.7	11.60	7.7
Ca	2.09	1.96	11.0	1.39	3.8
Mg	0.62	0.59	8.0	0.32	6.0
	(μgg^{-1})	(μgg^{-1})		(μgg^{-1})	
Fe	300	302	17.8	188	11.0
Mn	91	96	13.6	106	9.0
B	33	32	4.6	18	8.7
Zn	25	25	22.4	43	12.3
Cu	12	12	11.4	5	7.4
Mo	—	0.2	52.9	1	8.5

²N = 6 for each element of the NBS orchard leaves (NBS Standard Reference Material 1571) available from the National Bureau of Standards.

³N = 5 for each element of the first plant of experiment I for investigator C.

this mix is probably a result of root absorption of these elements directly from the peat-lite mix.

The overall content of essential elements in the leaf tissue was in the range normally considered sufficient for greenhouse (4, 6, 7, 8) and of field-grown lettuce (12, 14). The extremely rapid growth of the plants in these and previous studies (13) probably accounts for the fact that the concentrations of all the essential elements except K were in the low part of the sufficient range.

Although the nutrient status of the lettuce was in the normal range for good yielding lettuce, there was a high degree of variability in tissue elemental concentration. This is consistent with the observation that most plants do not respond to changes in tissue elemental content as long as the concentration remains within an adequate range (5, 13, 25, 28, 30). The variability for the 20 elements examined was such that no single element could be used as an index of overall nutrient status or as an adequate explanation for the variation in yield reported previously (13). Despite the magnitude of the differences in tissue composition obtained, there was no observable effect on yield (13). The tissue concentration of each element, or in some cases group of elements, seemed to be governed by a different set of factors. Examples of elemental tissue differences occurred at all levels among: 1) investigators; 2) experiments; 3) plants; and 4) subsamples.

Detailed data on growth responses on lettuce obtained by investigators A, C, and D are described by Hammer et al. (13) and that for investigator B by Krizek and Ormrod (20). Comparative data on elemental content of lettuce grown under different far-red and infrared levels are described by Krizek and Berry (19).

This study demonstrates the usefulness of multi-element analyses in the overall evaluation of growth systems and provides a comparative basis for future interlaboratory studies in plant nutrition under controlled environments.

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